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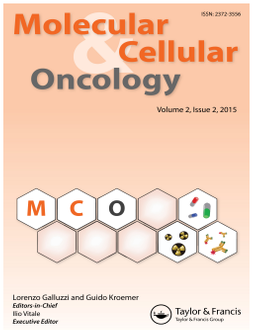

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p53 isoforms change p53 paradigm

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Although p53 defines cellular responses to cancer treatment it is not clear how p53 can be used to control cell fate outcome. Data demonstrate that so-called p53 does not exist as a single protein, but is in fact a group of p53 protein isoforms whose expression can be manipulated to control the cellular response to treatment.

TP53 is the most frequently mutated gene in human cancers and is thus one of the most studied (72,500 publications, pubmed August 2014). For 25 years it was thought that human *TP53* expressed only one protein, p53. In 2005, we reported that the human *TP53* gene expresses at least 12 p53 proteins (p53 isoforms) as a result of alternative splicing, alternative initiation of translation, and alternative promoter usage.[1] Each p53 isoform contains distinct protein domains. These p53 isoforms were also identified in mouse, zebrafish, and *Drosophila*, suggesting that they play important physiological roles since their expression is conserved through evolution. In normal tissue the human *TP53* gene expresses the following isoforms in a tissue-dependent manner: p53α (canonical p53), p53β, p53γ, Δ40p53α, Δ40p53β, Δ40p53γ, Δ133p53α, Δ133p53β, Δ133p53γ, Δ160p53α, Δ160p53β, and Δ160p53γ. Only p53α is expressed in every tissue. However, p53α is never expressed alone; rather, several p53 isoforms are always co-expressed with p53α in normal human tissue.[1] We and others have analyzed p53 isoform expression in a large panel of cancer cell lines and normal cells derived from diverse tissue origins. None of the normal and cancer cell lines (epithelial or fibroblast) expressed only canonical p53 (p53α) and p53α was always co-expressed with several p53 protein isoforms at the cellular level.[1-7]

A large body of evidence has now demonstrated that any changes in cell homeostasis activate a cellular response dependent on p53. Therefore, any anti-cancer drugs or treatments that affect cell homeostasis directly or indirectly activate p53 and trigger a p53-dependent cell response. In other words, the expression of wild-type (WT) TP53 gene determines whether a cell is going to survive, senesce, proliferate, differentiate, migrate, or die in response to cancer treatment. However, what is the so-called p53 protein? Is p53 only one protein (p53α) or a group of p53 isoforms? Which protein(s) encoded by the TP53 gene have the biological and biochemical activities attributed to p53? These questions are of paramount importance because the answers will have a profound impact on the treatment of cancer patients.

In a recently published article entitled "Modulation of p53β and p53γ expression by regulating the alternative splicing of TP53 gene modifies cellular response,"[8] we investigated whether p53 is a single protein, p53α, or a group of p53 protein isoforms including p53α. We determined that endogenous p53β and p53γ protein expression can be induced by manipulating the alternative splicing process. To achieve this, we treated a panel of WT TP53 cell lines with a novel specific inhibitor of Cdc2-like kinases, TG003. Cdc2-like kinases regulate some alternative splicing pre-mRNA processes by phosphorylating particular splicing factors such as SRSF1 and SRSF3. Importantly, inhibition of Cdc2-like kinases does not abolish all alternative splicing events.

We determined that inhibition of Cdc2-like kinases by TG003 or the knockdown of *SRSF1* promotes the inclusion of TP53 exons 9β/9γ and induces p53β and p53γ protein expression. Using siRNA that specifically targeted TP53 exons 9β/9γ with no effect on p53α expression, we established that endogenous p53β and p53γ inhibit cell proliferation by promoting cell death in MCF7 cells grown under standard culture condition. Conversely, by combining TG003 and siRNA targeting specifically TP53 exons 9β/9γ with no effect on p53α expression, we showed that endogenous p53β and p53γ proteins promote proliferation of MCF7 cells upon inhibition of Cdc2-like kinases by TG003. Therefore, p53β and p53γ have dual activities, promoting either death or proliferation of WT TP53 cells depending on the cellular context. Mechanistically, p53β and p53γ form stable protein complexes with p53α on the DNA of p53-responsive promoters such that oligomers composed of p53β and p53α and oligomers composed of p53γ and p53α regulate expression of different p53-responsive genes. The opposite activities of p53β and p53γ isoforms observed in non-treated and TG003-treated cells may reflect the effect of TG003 on both the expression and

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post-translational modifications of p53 isoforms, which might alter their oligomerization abilities.

Our data demonstrate that manipulation of endogenous p53β and p53γ protein expression using a splicing factor inhibitor and/or siRNA targeting specifically TP53 exons 9β/9γ, allows us to trigger different p53-mediated cell responses in a single cell line in a controlled way. This is consistent with previous data; we and others have previously shown that Δ40p53α and Δ133p53α oligomerize with p53α and regulate the p53-mediated cell response.[7]

For the past 10 years, we and others have investigated whether p53 is a single protein, p53α, or a group of p53 isoforms using different animal models (zebrafish, *Drosophila*, mouse) and different normal and cancer cell lines derived from distinct human tissues. Irrespective of the cell lines or animal models used, all of the data consistently indicate that the cell fate decision in response to damage or cell signals is defined by the p53 isoforms.[6-10] Therefore, we can now assert that the protein generally called p53 is NOT a single protein, p53α, but is in fact an ensemble of different oligomers, each composed of distinct p53 protein isoforms. Each oligomer has a different intrinsic transcriptional activity and promoter specificity. Hence, the p53-mediated cell response would be defined by the sum of the activities of each p53 isoform oligomer. This would explain why manipulation of the cellular composition of p53 isoforms by small molecules can trigger opposite p53-dependent cell fate outcomes (repair/survival/proliferation or cell death) in the same cell type in response to the same treatment. It is thus imperative to decipher the mechanism of p53-isoform mediated cell fate decisions for efficient clinical application of anticancer drugs including the new p53-targeting drugs such as Nutlin, Prima, and Rita.

In light of the literature discussed here, p53 isoforms offer novel exciting perspectives in basic and translational research that I am convinced will revolutionize cancer treatment, improving patient quality of life and survival.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

References

1. Bourdon JC, Fernandes K, Murray-Zmijewski F, Liu G, Diot A, Xirodimas DP, Saville MK, Lane DP. p53 isoforms can regulate p53 transcriptional activity. Genes Dev 2005; 19:2122-37; PMID:16131611; http://dx.doi.org/10.1101/gad.1339905
2. Aoubala M, Murray-Zmijewski F, Khoury MP, Fernandes K, Perrier S, Bernard H, Prats AC, Lane DP, Bourdon JC. p53 directly transactivates Delta133p53alpha, regulating cell fate outcome in response to DNA damage. Cell Death Differ 2011; 18:248-58; PMID:20689555; http://dx.doi.org/10.1038/cdd.2010.91
3. Terrier O, Marcel V, Cartet G, Lane DP, Lina B, Rosa-Calatrava M, Bourdon JC. Influenza A viruses control expression of proviral human p53 isoforms p53beta and Delta133p53alpha. J Virol 2012; 86:8452-60; PMID:22647703; http://dx.doi.org/10.1128/JVI.07143-11
4. Avery-Kiejda KA, Zhang XD, Adams LJ, Scott RJ, Vojtesek B, Lane DP, Hersey P. Small molecular weight variants of p53 are expressed in human melanoma cells and are induced by the DNA-damaging agent cisplatin. Clin Cancer Res 2008; 14:1659-68; PMID:18310316; http://dx.doi.org/10.1158/1078-0432.CCR-07-1422
5. Takahashi R, Markovic SN, Scrable HJ. Dominant effects of Delta40p53 on p53 function and melanoma cell fate. J Invest Dermatol 2013; 134:791-800; PMID:24037342; http://dx.doi.org/10.1038/jid.2013.391
6. Mondal AM, Horikawa I, Pine SR, Fujita K, Morgan KM, Vera E, Mazur SJ, Appella E, Vojtesek B, Blasco MA, Lane DP, Harris CC. p53 isoforms regulate aging- and tumor-associated replicative senescence in T lymphocytes. J Clin Invest 2013; 123:5247-57; PMID:24231352; http://dx.doi.org/10.1172/JCI70355
7. Surget S, Khoury MP, Bourdon JC. Uncovering the role of p53 splice variants in human malignancy: a clinical perspective. Onco Targets Ther 2013; 7:57-68; PMID:24379683
8. Marcel V, Fernandes K, Terrier O, Lane DP, Bourdon JC. Modulation of p53beta and p53gamma expression by regulating the alternative splicing of TP53 gene modifies cellular response. Cell Death Differ 2014; 21:1377-87; PMID:24926616; http://dx.doi.org/10.1038/cdd.2014.73
9. Dichtel-Danjoy ML, Ma D, Dourlen P, Chatelain G, Napoletano F, Robin M, Corbet M, Levet C, Hafsi H, Hainaut P, Ryoo HD, Bourdon JC, Mollereau B. Drosophila p53 isoforms differentially regulate apoptosis and apoptosis-induced proliferation. Cell Death Differ 2013; 20:108-16; PMID:22898807; http://dx.doi.org/10.1038/cdd.2012.100
10. Ou Z, Yin L, Chang C, Peng J, Chen J. Protein interaction between p53 and Delta113p53 is required for the anti-apoptotic function of Delta113p53. J Genet Genomics 2014; 41:53-62; PMID:24576456; http://dx.doi.org/10.1016/j.jgg.2014.01.001